Annual Report for

Navy Contract N00014-93-1-1263

Response of the Lungs to Low Frequency Underwater Sound

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The purpose of this report is to annotate the slides from our research report given at the Summary Meeting on the Evidence to Date, 29 June 1994, on the Effects of Low Frequency Water-Borne Sound on Divers at the Naval Submarine Base New London, Groton, Connecticut.

SLIDE 1 - Response of the Lungs to Low Frequency Underwater Sound

The animal research was performed in conjunction with the Division of Animal Resources at Emory University. The measurements on human subjects were performed at the Ocean Simulation Facility in Panama City, Florida with the help of the Navy Experimental Diving Unit.

SLIDE 2 - Because of their high compliance...

Because of their high compliance, the lungs are probably more vulnerable to damage by high intensity, low frequency underwater sound than any other part of the human anatomy. The objective of this research was to study the effects of low frequency (50 - 1200 Hz) underwater sound on the lungs and to determine safe exposure conditions for swimmer and divers. In order to assess the risk, it is necessary to have a complete understanding of the vibrational response of the lungs as well as knowledge of the potential damage mechanism. It is of principle importance to determine the resonance frequencies of the lungs, because for a given low frequency excitation, the vibration of the lungs (and hence the risk of damage) will be highest at resonance. These resonances must be measured underwater, not in air, since the resonance frequencies may be affected by the additional fluid loading underwater. The lung resonance frequencies may also be a function of depth and the air volume in the lungs.

The original research plan to characterize the vibrational response involved measuring the lung resonance of an animal model as well as humans. The animal model would later be used to attempt to establish damage thresholds. A full size animal model was needed since the scaling of the mechanics and physiology of the lung is not well understood. The domestic pig was chosen as it reasonable represents human physiology and is commonly used.

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May 10, 1995

REPLY TO: E-25-W28

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SUBJECT: Annual Performance Report
Project Director: P.H. Rogers
Contract No.:N00014-93-1-1263
"RESPONSE OF THE LUNGS TO LOW FREQUENCY UNDERWATER SOUND"
Period Covered:930901 - 940831

The subject report is forwarded in conformance with the contract/grant specifications.

Should you have any questions or comments regarding this report(s), please contact the Project Director or the undersigned at 404-894-4764.

/tw

Sincerely,

Wanda W. Simon

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Research Reports Coordinator

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Response of the Lungs to Low Frequency Underwater Sound

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Research Performed in Conjunction with

Division of Animal Resources
Emory University

Navy Experimental Diving Unit

Because of their high compliance, the lungs may be susceptible to injury from low frequency underwater sound.

The goal of this project is to measure the vibrational response of the lungs in the range from 50-1200 Hz using a sound exposure level of 130 dB.

1. Measure Lung Resonance in Animal Model (domestic pigs)

Small Tank (500 gal)	- Head out	Developmental
	- Head underwater	"
Big Tank (32,000 gal)	- Head out	Research
	- Immersed to 7 ft.	"

2. Measure Lung Resonance in Humans

Big Tank	- Head out
	- Immersed to 10 ft.
Wet Pot	- Immersed to 5, 33, 66, and 100 ft.

The plan was to develop the measurement technique on the animal model in a small tank (500 gal), both with the head of the animal above the water surface and with the head underwater. Quantitatively, the data collected during the small tank developmental phase were compromised due to the relative size of the lung volume to the tank volume. Therefore, measurements were also to be made in a big acoustic tank (32,000 gal), both head out and immersed to mid depth (~7 ft). Unfortunately, the measurements on the animal model in the big tank were never made.

Next, the plan was to make lung resonance measurements on humans, head out and immersed, in the same big acoustic tank under the same conditions for direct comparison. Then measurements were to be made in the Ocean Simulation Facility (wet pot) to examine the effects of depth on lung response. Unfortunately, again, this plan was not followed. Measurements were made on three subjects with their head out in the big tank, and measurements were made on six subjects, head out and immersed to 10 ft, in the test pool (30 ft x 15 ft x 15 ft) on the OSF site.

SLIDE 3 - Proposed Time Schedule

The proposed time schedule lists the original plan for developmental effort and research effort for:

1. Lung resonance measurements on the animal model (Pig NIVAMS Study),
2. Lung damage threshold study on the animal model (Pig Lung Damage Study),
3. Lung resonance measurements on humans (Human NIVAMS Study),
4. Analytical modeling of the lung resonance in humans and the animal model (Analytical Study), and
5. Recommendations based on the research results (Recommendations).

Proposed Time Schedule

[-------> Developmental Effort ————> Research Effort]

	JAN	FEB	MAR	APR	MAY	JUN	JUL	AUG	SEP	FY-95
1. PIG NIVAMS STUDY										
Small Tank - Head out										
- Head under										
Big Tank - Shallow										
- Deep (7ft)										
Data Runs										?
(24 pigs 1 1/2 pigs/week)†										
2. PIG LUNG DAMAGE STUDY (24 pigs 1 1/2 pigs/week)†										
Shallow										?
Deep (7 ft)										?
Wet Pot										
3. HUMAN NIVAMS STUDY (2 subjects/week)										
Small Tank										
Big Tank										?
Wet Pot					<-->					<-->
4. ANALYTICAL STUDY										
Model Development										
Validation & Data Analysis										
5. RECOMMENDATIONS						X			X	?

† Pigs used in the NIVAMS study are also used in the intense sound, lung damage study

This presentation summarizes the work completed from this ambitious plan and represents the recommendations due at the end of June.

SLIDE 4 - NIVAMS

The lung resonance was measured using the NIVAMS (Non-Invasive Vibration Amplitude Measurement System). The NIVAMS uses ultrasound to measure, *in vivo*, the vibration of tissue and organs induced by low frequency underwater sound. The advantages of the NIVAMS are that it is non invasive in that no surgery is required to make the measurement and it is non intrusive in that the vibrational response is unaltered by the measurement process.

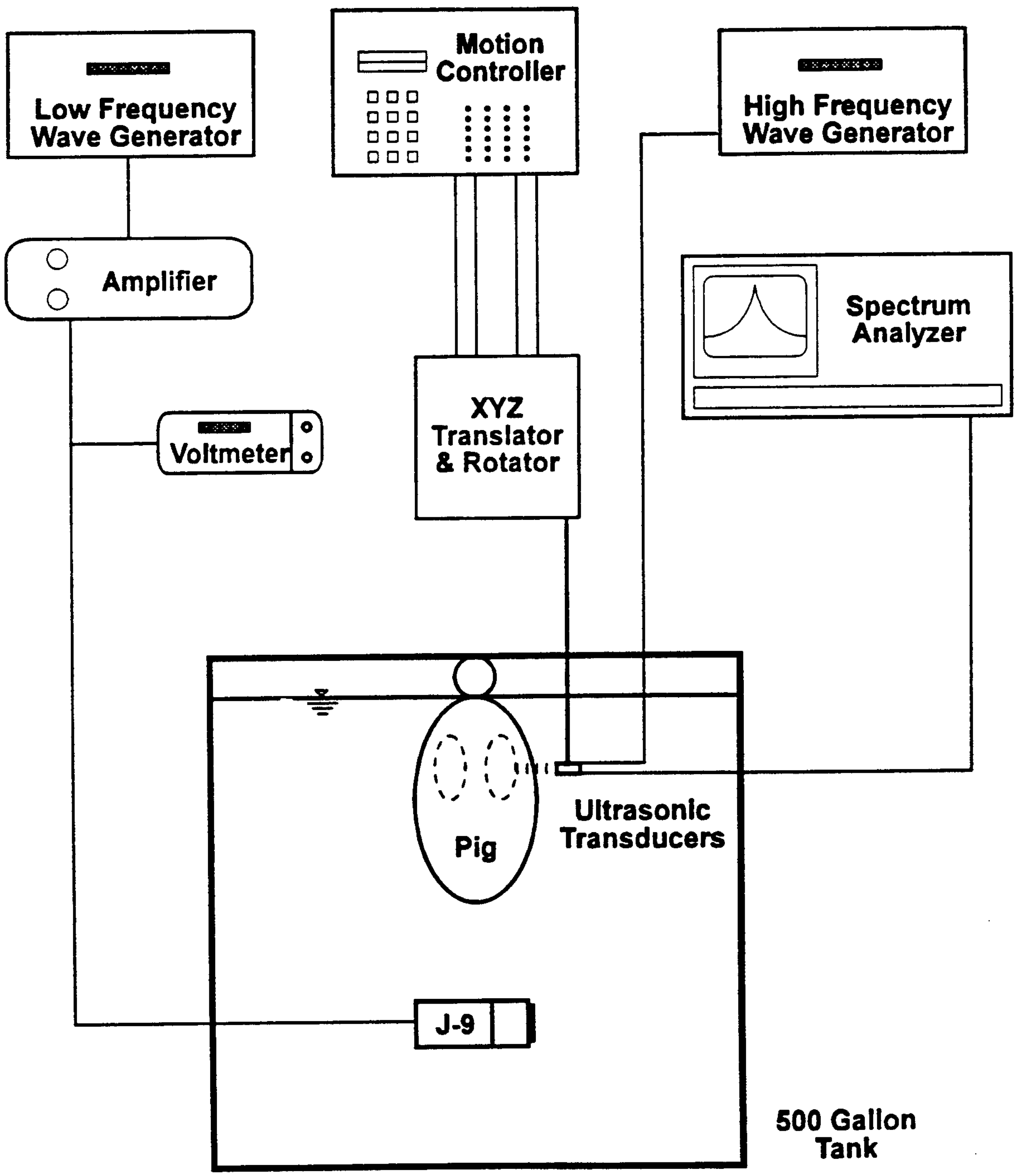
The NIVAMS consisted of three subsystems - the underwater sound generator, the ultrasonic transmitter and receiver, and the transducer positioner. The low frequency underwater sound was created by a wave generator connected to an underwater transducer (J-9). The transmitted 1 MHz ultrasound was created by a high frequency wave generator connected to an ultrasonic transducer. A second identical transducer was used as a receiver, whose signal is viewed on a spectrum analyzer (continuous wave, frequency domain) or an oscilloscope (pulse-echo, time domain). The two transducers were mounted in a fixture that was positioned with an XYZ translator and rotator for the animal study. For the human study, the transducers were positioned by the subject with manual translators. The transducers were positioned to view the surface of the lung through the intercostal space.

SLIDE 5 - NIVAMS Pulse-Echo Signals

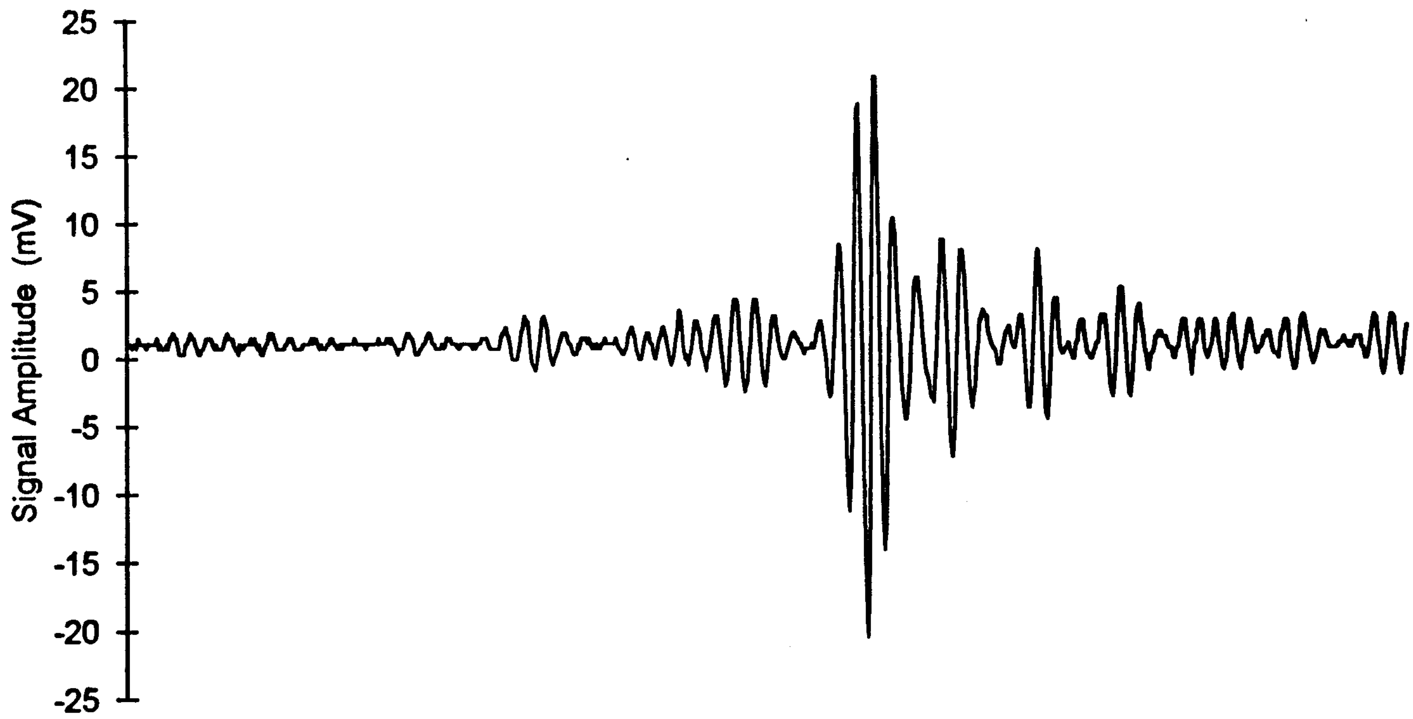
The pulse-echo NIVAMS mode was used to locate and identify organs in the body of the subject. The transmitted signal consisted of single 1 MHz sine wave pulses at a 1 KHz repetition rate. This ultrasound traveling through the body is reflected at tissue interfaces. The relative difference in characteristic impedance on the two sides of the interface determines the amplitude of the reflection. Since the characteristic impedance of the air filled lung is much different than that of the tissue in the intercostal space, the lung surface returns a large echo. The transducers

NIVAMS

(Non-Invasive Vibration Amplitude Measurement System)



NIVAMS Pulse-Echo Signals: Time Domain



were positioned to maximize the reflected signal from the lung surface and minimize all other signals. The rigid rib also returns a substantial echo, but can be discriminated from the compliant lung, as the one is 180 degrees out of phase from the other.

SLIDE 6 - NIVAMS Continuous Wave Signals

The continuous wave NIVAMS mode was used to measure the amplitude of vibration of the reflecting surface. The reflected ultrasound is phase modulated by the moving interface. In the frequency domain, some of the energy from the 1 MHz transmitted signal is shifted to side bands at the ultrasound frequency (1 MHz) plus and minus the underwater sound frequency (shown at 50 Hz). The amplitude of vibration can be calculated from the relative amplitude of the side bands to the center frequency.

SLIDE 7 - Vibrational Response of Porcine Lungs

By varying the underwater sound stimulus frequency in discrete steps , measurements were made to generate a frequency response curve for the lungs. Slide 7 shows that the measurements were repeatable for a given subject on a given day.

SLIDE 8 - Water Particle Motion

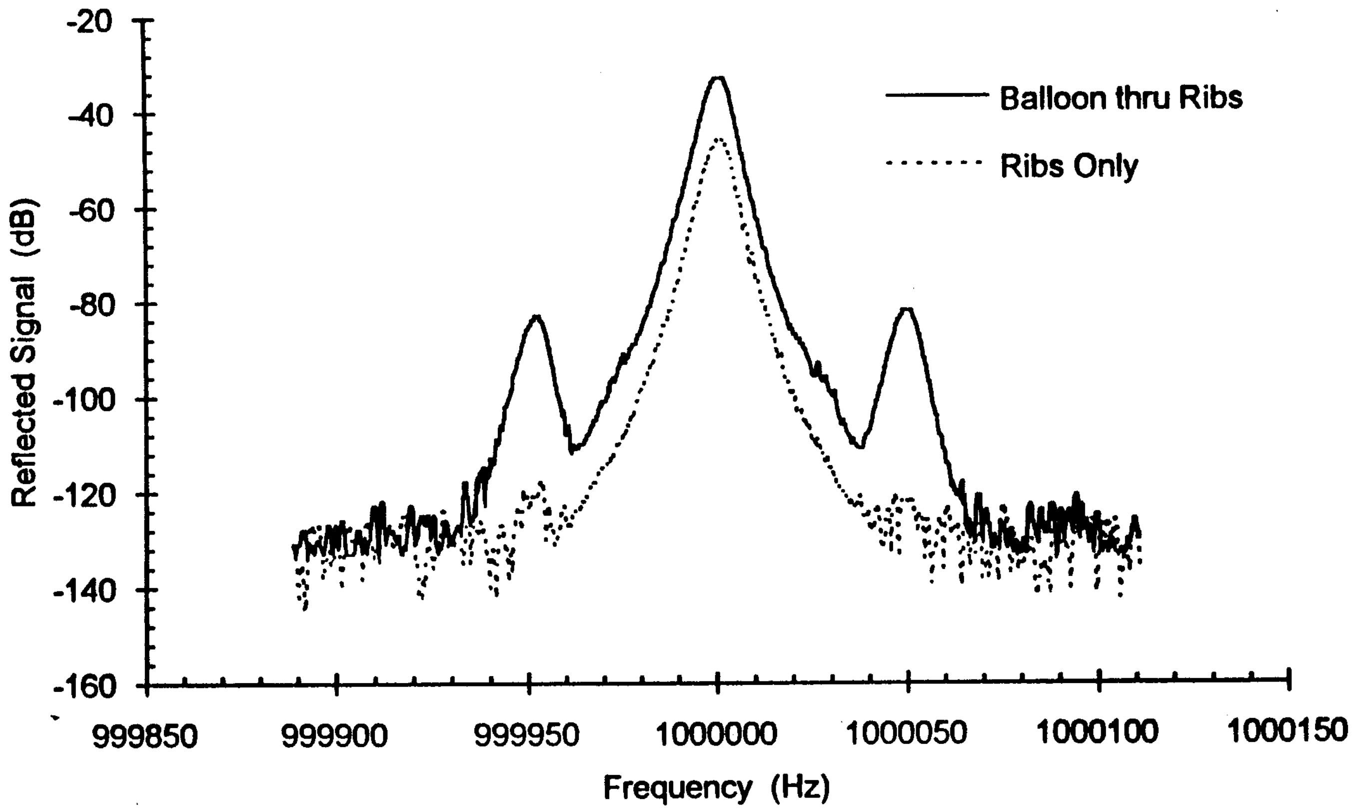
The acoustic particle motion in the small tank caused by the low frequency sound stimulus was calculated from measured pressure data. The amplitude of the water was found to be much less than the amplitude of the vibrational motion measured with the NIVAMS.

SLIDE 9 - Vibrational Response of Porcine Lungs: Diaphragm vs. Between Ribs

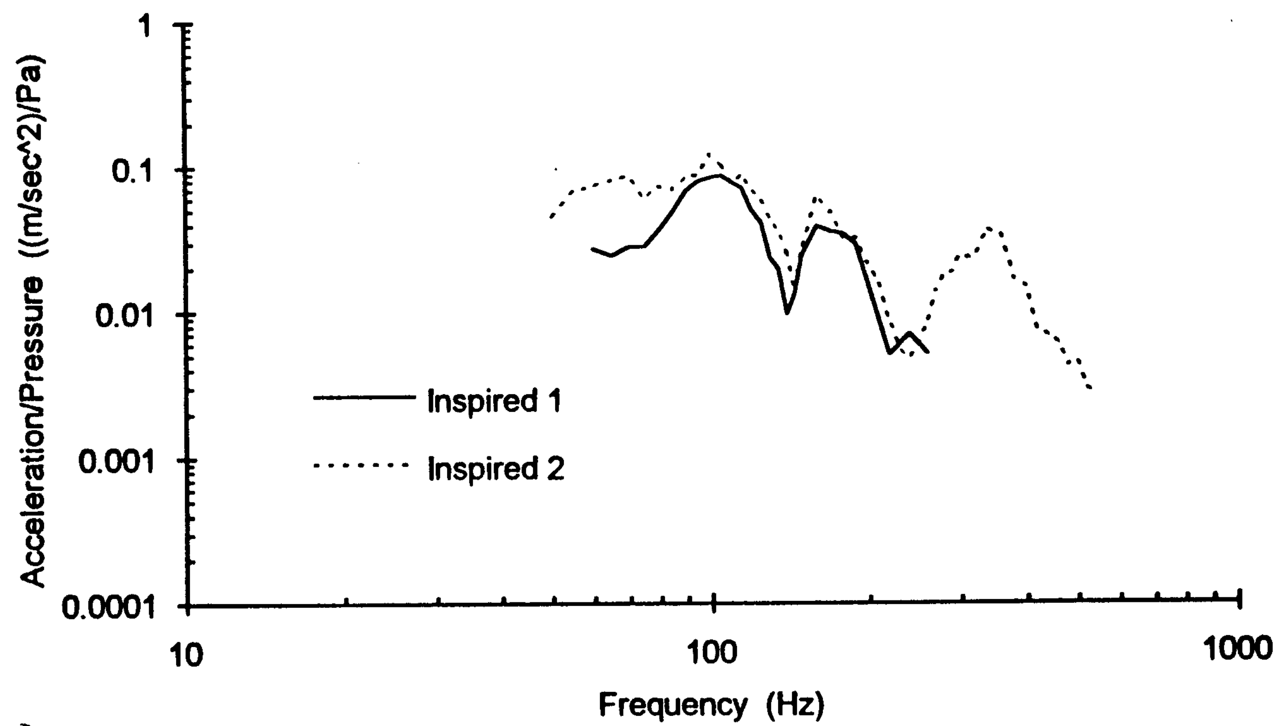
An attempt was made to look at the surface of the lung from a different orientation. The measured frequency response was similar for the two surfaces of the lung.

SLIDE 10 - Vibrational Response of Porcine Lungs: Alive vs. Deceased

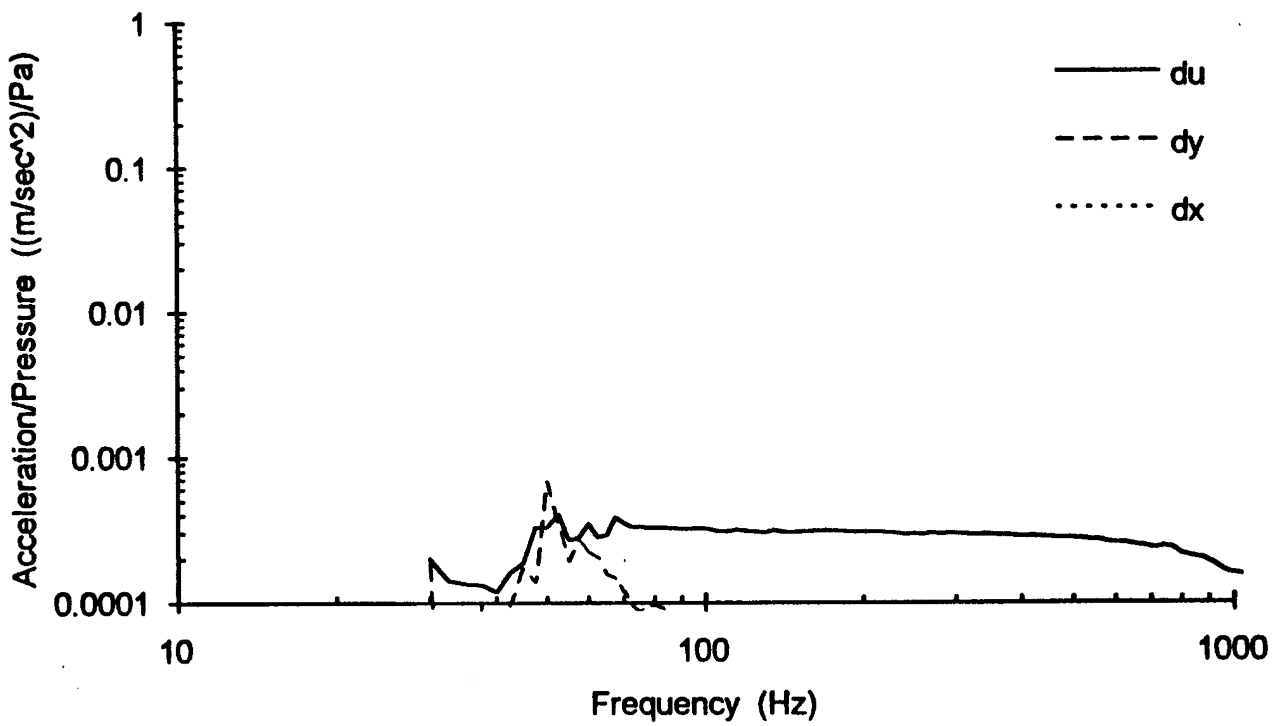
NIVAMS Continuous Wave Signals: Frequency Domain



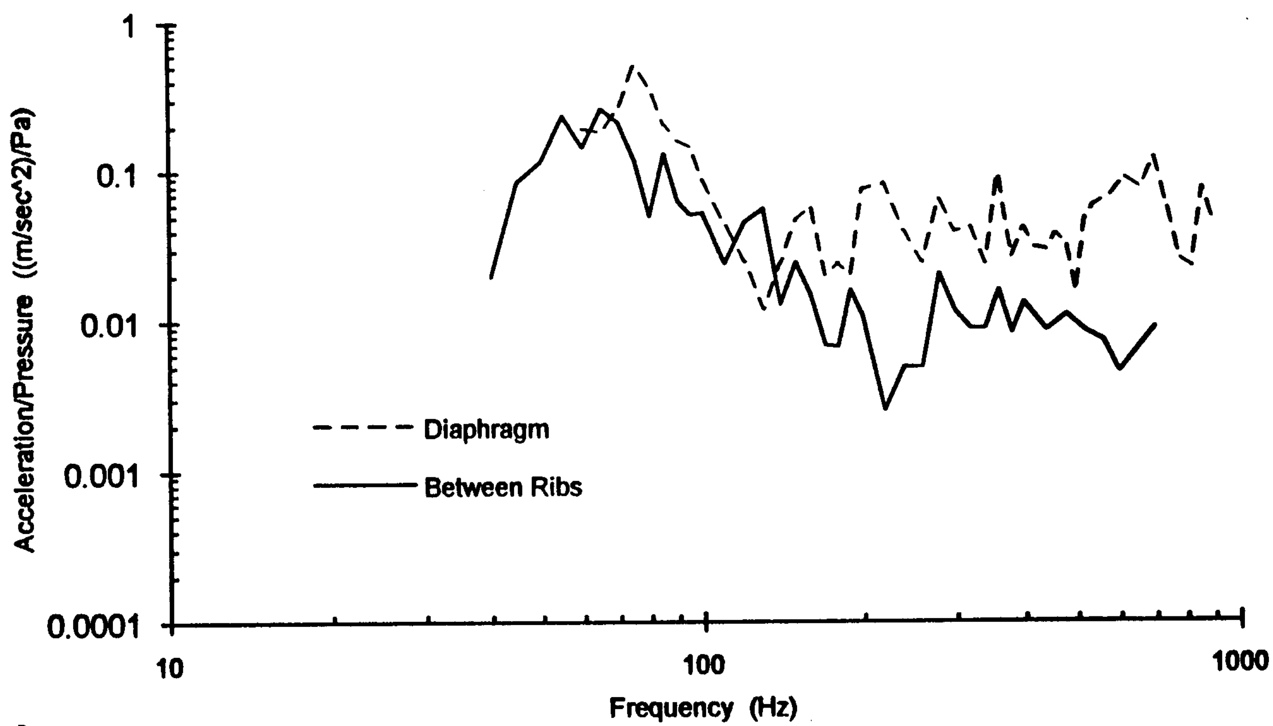
Vibrational Response of Porcine Lungs



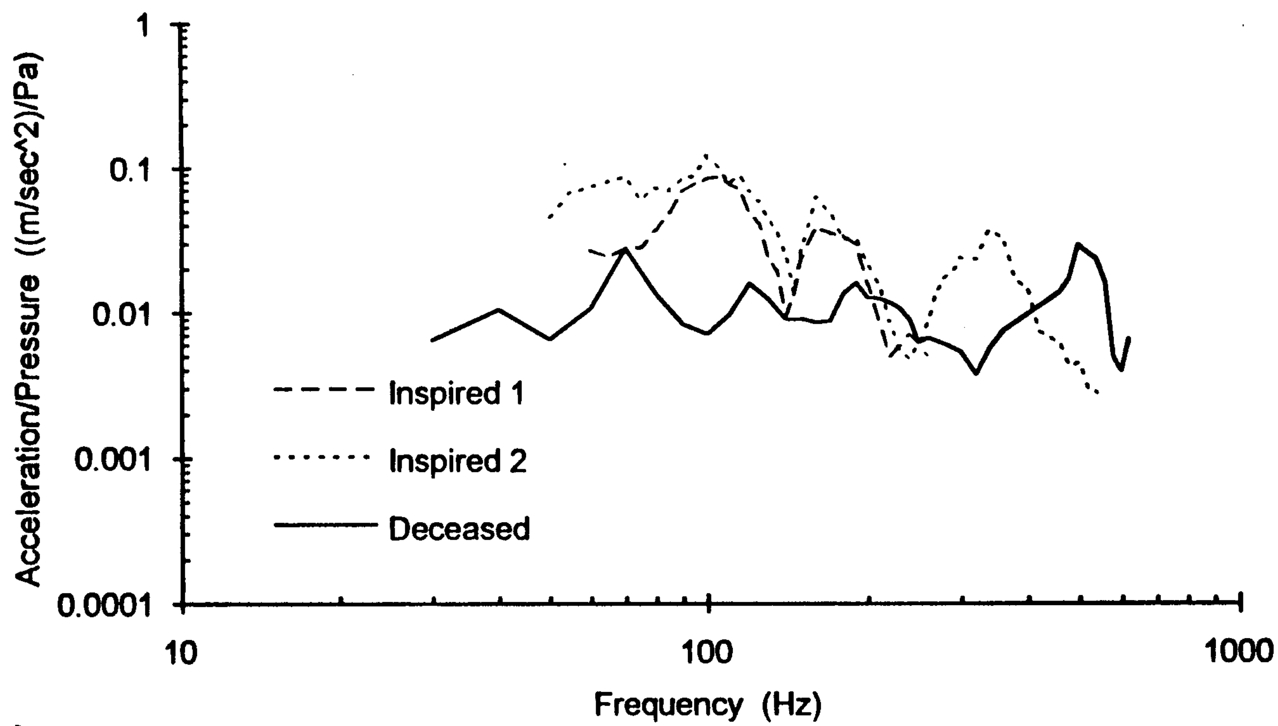
Water Particle Motion



Vibrational Response of Porcine Lungs
Diaphragm vs. Between Ribs



Vibrational Response of Porcine Lungs
Alive vs. Deceased



The frequency response was measured for an animal subject that had died during test preparation. This response was significantly different than that from live animals.

SLIDE 11 - Vibrational Response of Human Lungs: Lung vs. Rib

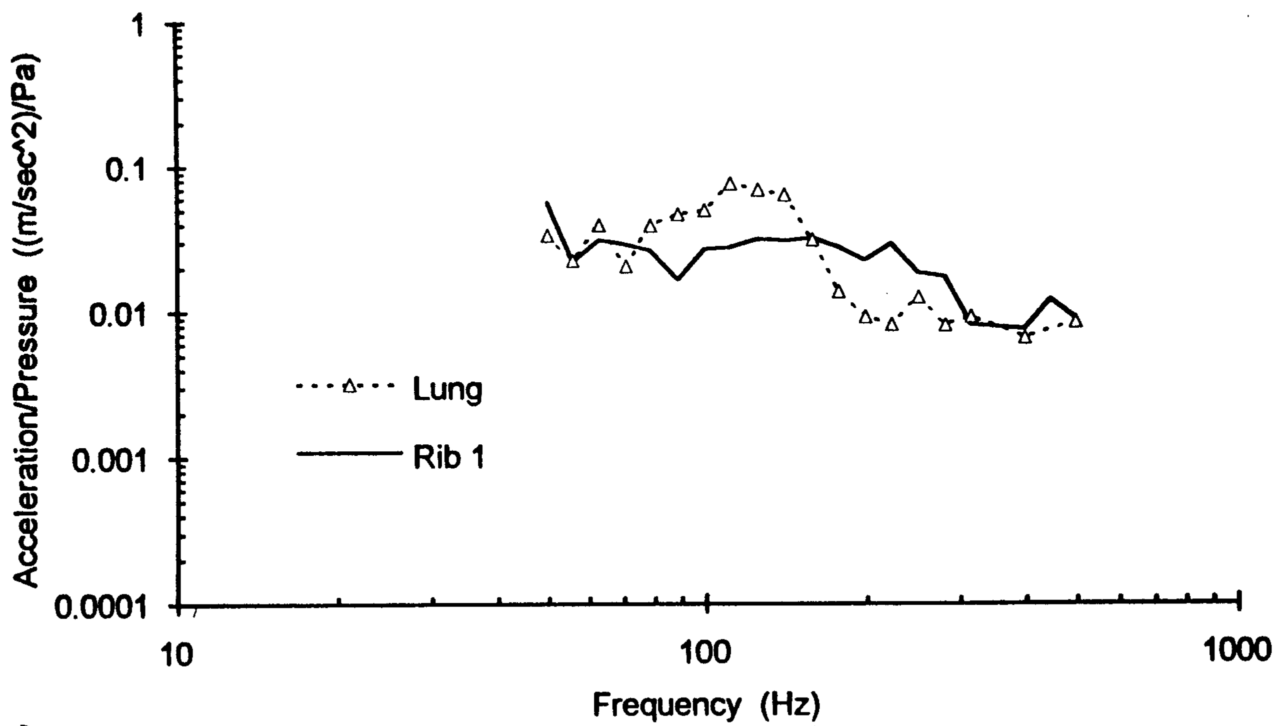
For one human subject, the vibrational response of a rib was measured to be different than the adjacent lung surface.

SLIDES 12 - 16 - Vibrational Response of Human Lungs: Head out vs. 10 ft. Under - Subjects A - E

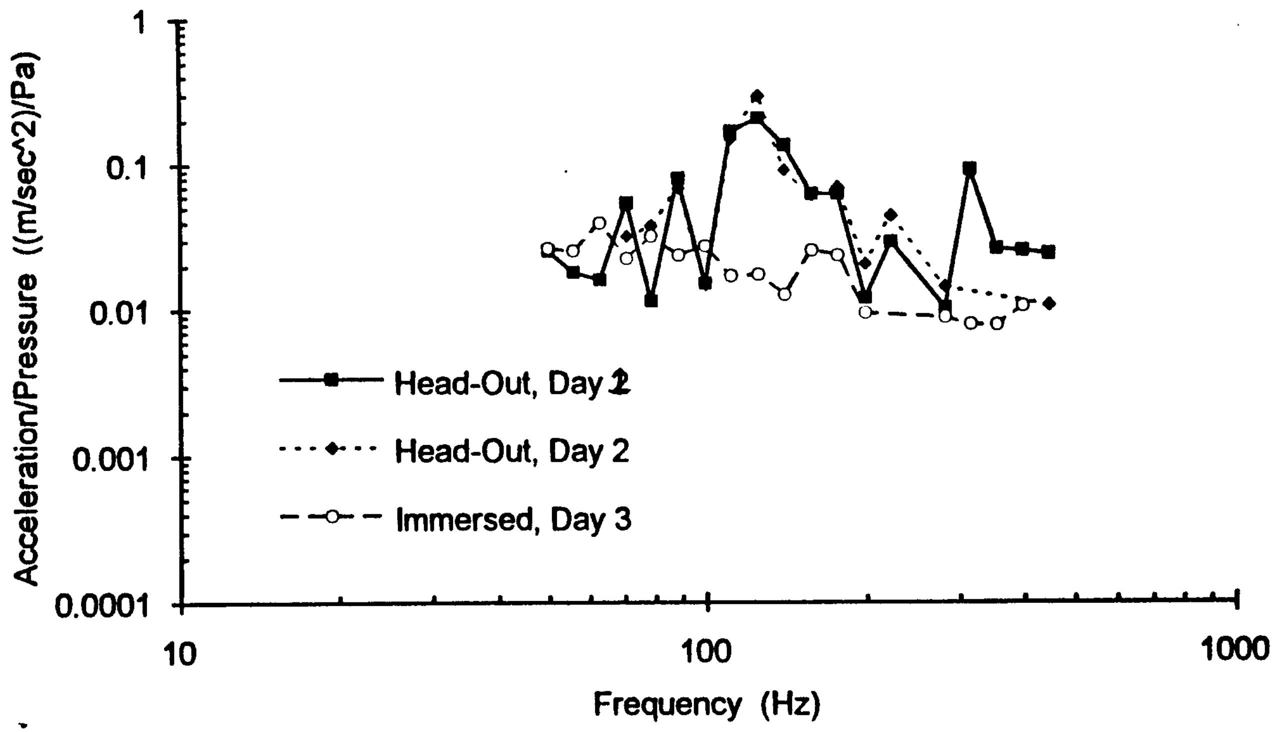
Lung resonance measurements using NIVAMS were made on a team of six divers in the test pool at NEDU during the week of May 23-27. Each diver was tested on two consecutive days under each of the following conditions: 1) Head-out, with a substernal depth of approximately 8.5 inches, and 2) Immersed to a substernal depth of approximately 10 feet. Vibration amplitudes were measured at discrete frequencies from 50 to 500 Hz in 1/6 octave steps, using a sound pressure level of not more than 130 dB (re: 1 μPa). While underwater, the subjects used a MK-20 diving rig. The diving rig was not used for the head-out study.

The filled symbol data was for the subject with their head-out. The general shapes of the curves were consistent, indicating a resonance in the 100-200 Hz range. This is probably not the fundamental lung resonance, however. The fundamental resonance was probably below the frequency range of this study. This secondary resonance between 100 and 200 Hz is consistent with the animal data. With the diver immersed in 10 feet of water, this resonance is reduced in amplitude, as shown by the open symbol data. This is a common feature for the data from all subjects: a resonance in the 100 to 200 Hz range head-out whose magnitude is reduced when the subject is underwater. A change in resonance frequency with depth is not obvious from the collected data. Data is shown for five of the six subjects. The subjects were instructed to hold their breath during the individual measurements to assure constant lung volume. The sixth

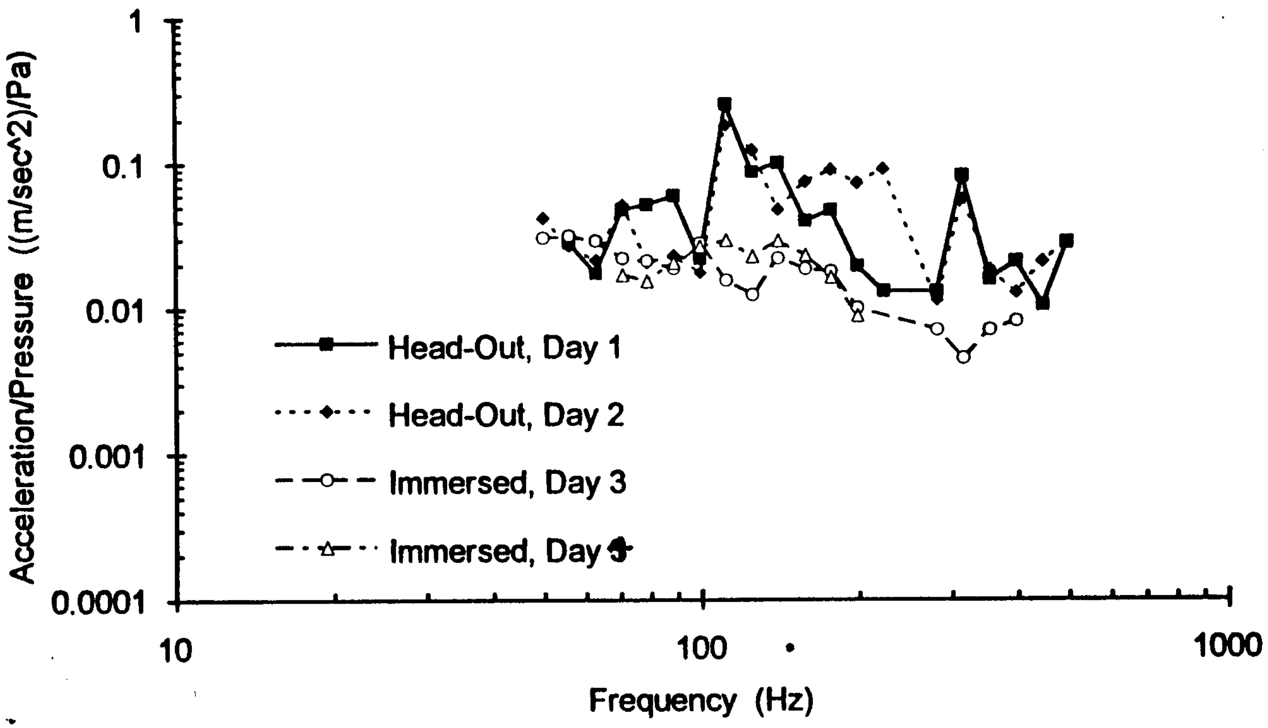
Vibrational Response of Human Lungs
Lung vs. Rib



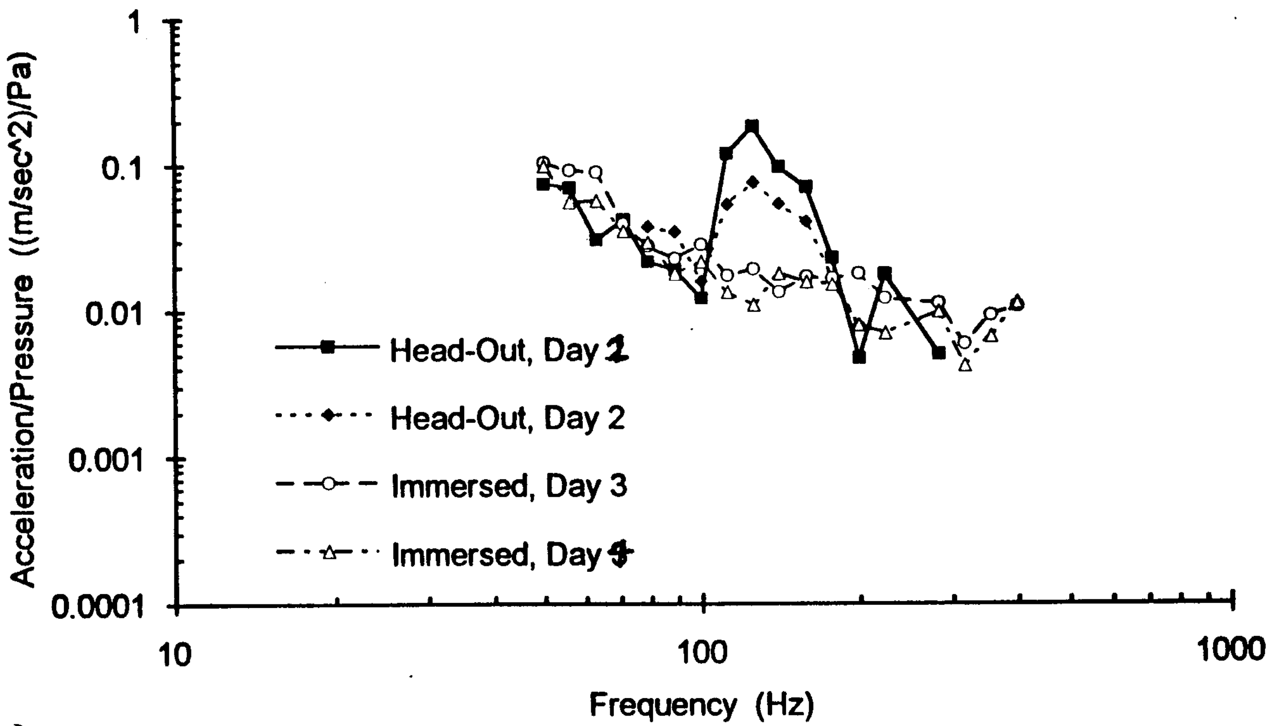
Vibrational Response of Human Lungs
Head out vs. 10 ft. Under
Subject A



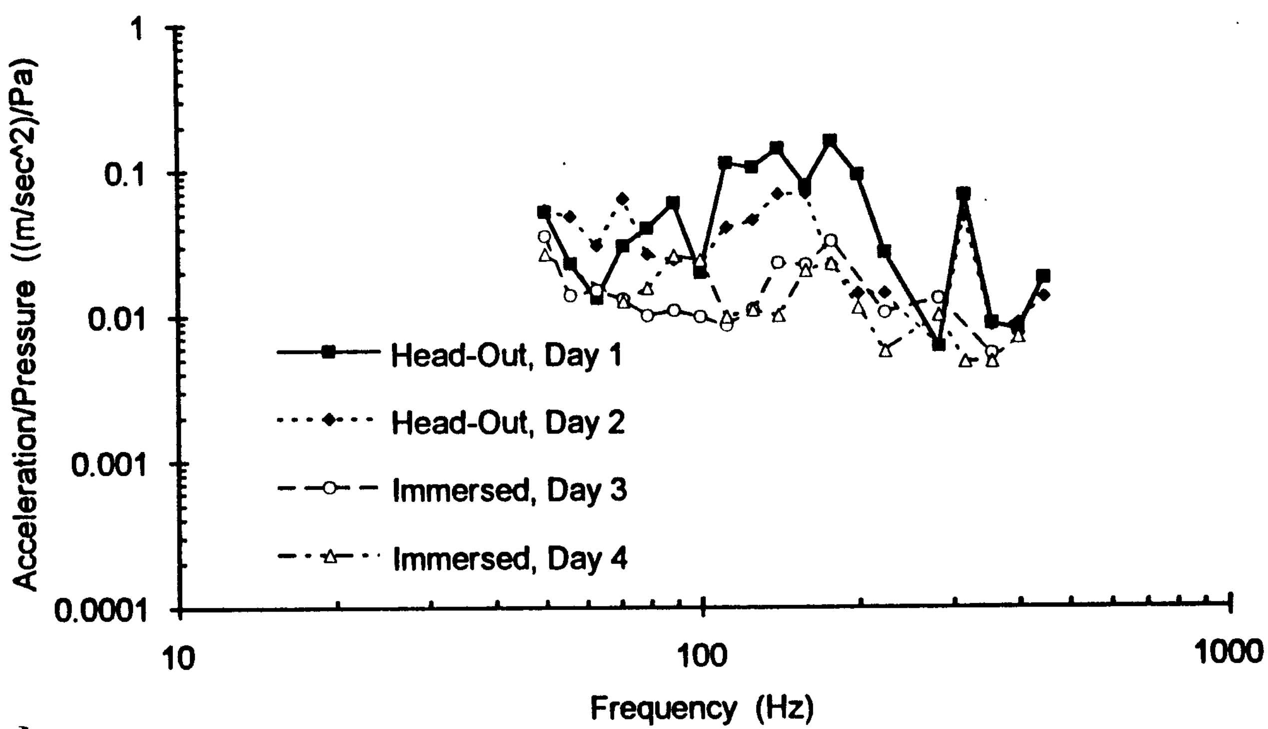
Vibrational Response of Human Lungs
Head out vs. 10 ft. Under
Subject B



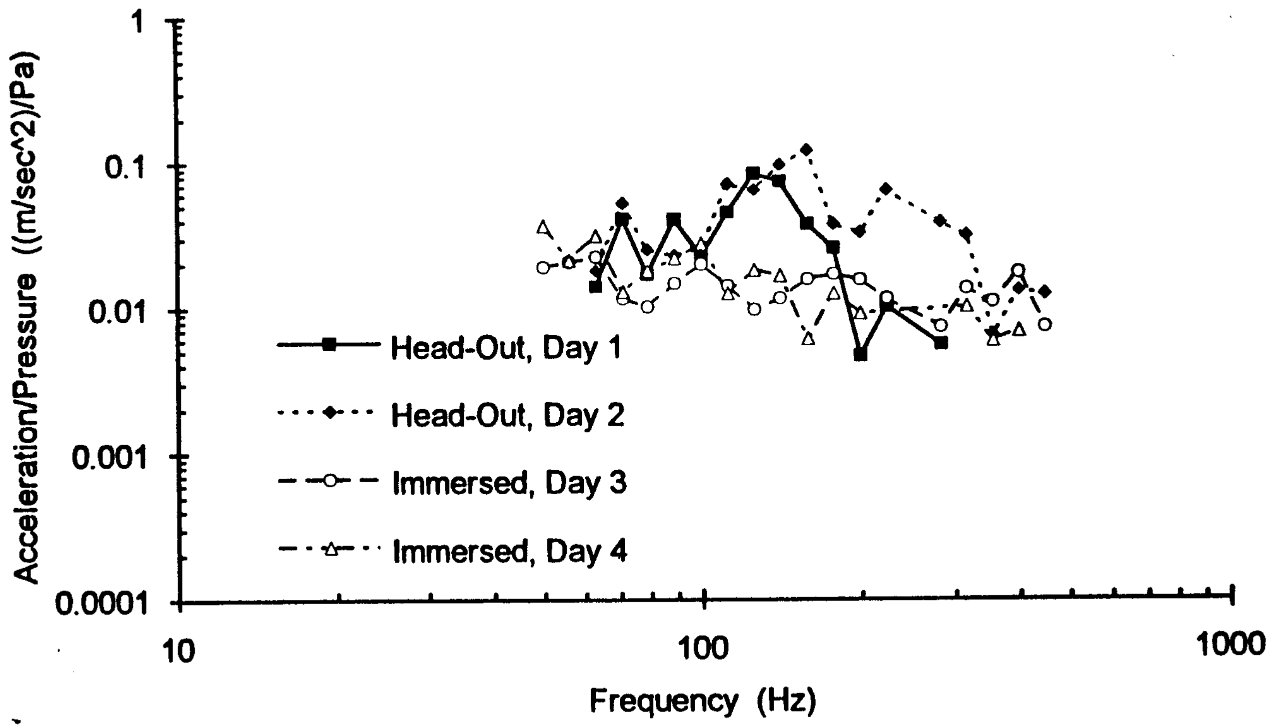
Vibrational Response of Human Lungs
Head out vs. 10 ft. Under
Subject C



Vibrational Response of Human Lungs Head out vs. 10 ft. Under Subject D



Vibrational Response of Human Lungs
Head out vs. 10 ft. Under
Subject E



subject did not do this (bubbles were noted during the underwater tests), so the data on subject F were rejected.

SLIDE 17 - Results from NIVAMS Study

The results from the NIVAMS study were:

1. The NIVAMS was able to measure the motion of the lungs in response to low frequency underwater sound.
2. The measured lung motion differed significantly from the acoustic particle motion.
3. The measured lung motion differed significantly from the motion of the overlying ribs.
4. The response from a freshly deceased animal differs significantly from a live animal.
5. For humans at the surface, there is a resonance in the 100 to 200 Hz range.
6. This resonance is reduced in amplitude when the subject is at a 10 foot depth.

SLIDE 18 - PUBA System

At the sponsors request, we attempted to determine if 170 dB underwater sound damaged the lungs of pigs. The test involved anesthetizing the animal and submerged it to middepth (6' 8") in our 32,000 gallon acoustic test tank. The test subject was then exposed to the high intensity, low frequency sound for 5 minute durations at discrete frequencies between 100 and 400 Hz. The subjects were then removed from the water, sacrificed, and necropsied to determine the effects of the exposure.

We developed an underwater breathing apparatus which was capable of maintaining an anesthetized pig. The Porcine Underwater Breathing Apparatus concept is fairly simple. The normal animal respirator was sealed in a pressure chamber. The pressure inside the chamber was regulated by an exhaust tube open at the depth of the animal's chest. Therefore, the pressure

Results from NIVAMS Study

1. The NIVAMS was able to measure the motion of the lungs in response to low frequency underwater sound.

2. The lung motion differed significantly from the acoustic particle motion.

3. The lung motion differed significantly from the motion of overlying ribs.

4. The response from a freshly deceased animal differs significantly from a live animal.

5. For humans at the surface, there is a resonance in the 100 to 200 Hz range.

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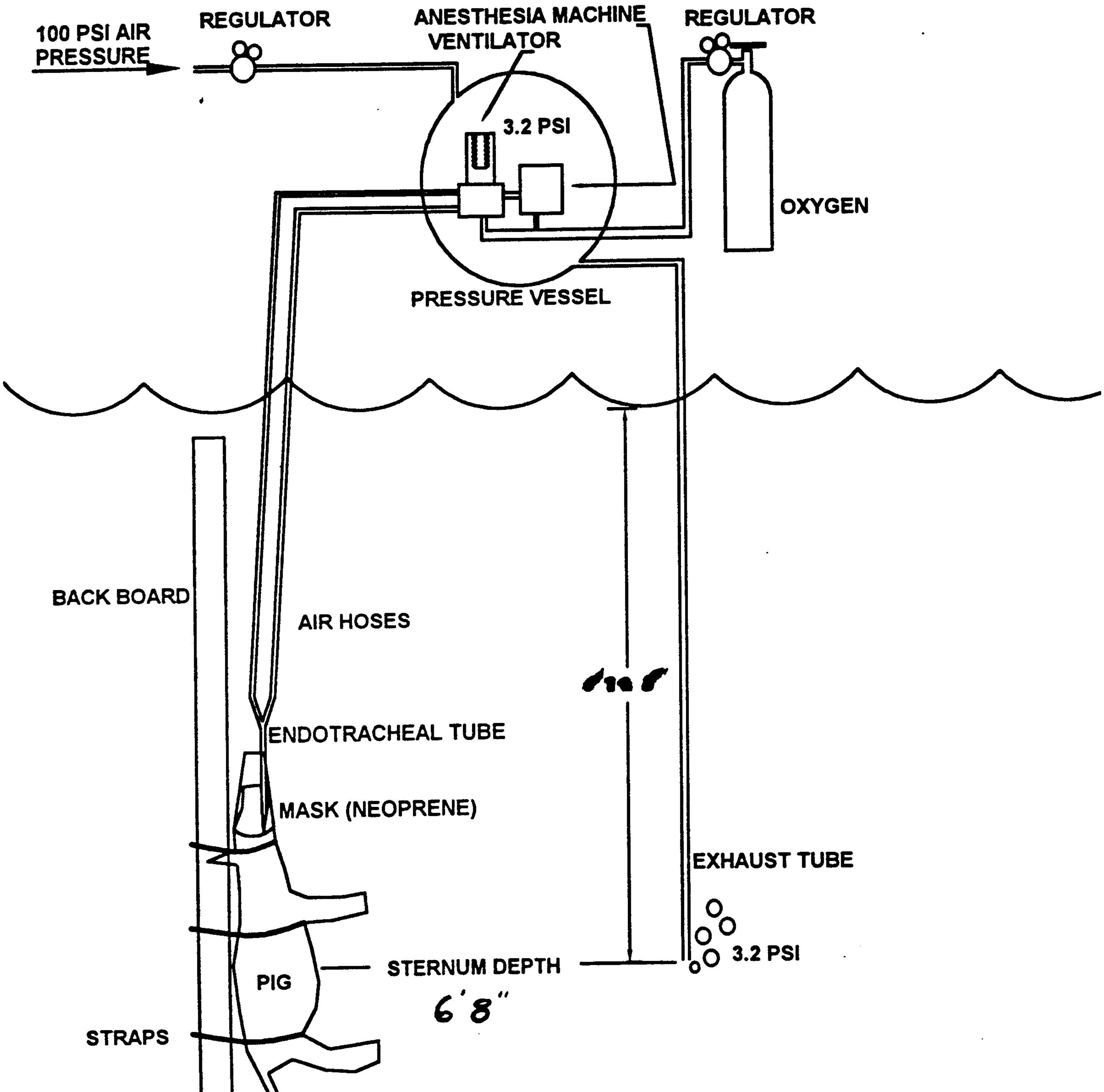
PUBA SYSTEM



100 PSI AIR PRESSURE
REGULATOR
ANESTHESIA MACHINE VENTILATOR
REGULATOR
3.2 PSI
OXYGEN
PRESSURE VESSEL
BACK BOARD
AIR HOSES
ENDOTRACHEAL TUBE
MASK (NEOPRENE)
EXHAUST TUBE
3.2 PSI
PIG
STERNUM DEPTH
6'8"
STRAPS

surrounding the ventilator is at the same ambient pressure as the lungs of the animal, and the airway pressure is kept relatively normal.

SLIDE 19 - Effects of Anesthetic Regimen and Underwater Sound on Cardiovascular Status, Acid-Base Balance and Pathological Changes in the Lungs of Pigs

The anesthetic regimen chosen and the ventilation management of the animal produced unexpected complications and had a strong effect on the outcome of some of the tests. Several animals suffered respiratory alkalosis due to improper ventilation management. Other animals have suffered hypotension, exacerbated by immersion in the water. Blood pressure management by eliminating the vasodilators given to the animals along with the initial sedatives and other measures helped minimize the hypotension. Both respiratory alkalosis and hypotension could have been responsible for the appearance of edema in the lungs of the animals. Unfortunately, damage to the lung tissue from sound exposure may also be identified by the appearance of interstitial or alveolar edema.

Three animals were exposed to underwater sound while under anesthesia using pentobarbital. During the exposure, the animals had normal cardiovascular and acid-base status. The lung tissue appeared normal after exposure.

SLIDE 20 - Sound Exposures

The animals were exposed to sound in a cylindrical water tank, 13 ft 4 in deep by 20 ft diameter. The animals were suspended in the center, above a USRD J-15-3 sound projector. The exposure sound pressure levels were estimated to be from 161 to 177 dB (re: 1 μPa) for the frequencies listed. The pressure levels were limited by the source strength and the configuration of the test tank.

SLIDE 21 - Results from Exposure Study

Effects of Anesthetic Regimen and Underwater Sound on Cardiovascular Status, Acid-Base Balance and Pathologic Changes in the Lungs of Pigs

Anesthetic Regime	N	Experimental Conditions [1]	Cardiovascular Status - Normal/Total	Acid-Base Status - Normal/Total	Lung Status - Normal/Total
Isoflurane	5	A	0 / 3 [2]	1 / 2 [2]	2 / 5 [3]
Isoflurane	2	B	1 / 2	1 / 2	1 / 2 [3]
Isoflurane	3	C	1 / 3	2 / 3	1 / 3 [3]
Pentobarbital	3	C	3 / 3	3 / 3	3 / 3

Notes:

[1] Experimental Condition A - Animals died while submerged or shortly after being prematurely removed from the tank. Not exposed to sound.

Experimental Condition B - Controls. Animals survived sham sound procedures at proper water depth in tank.

Experimental Condition C - Sound-exposed animals survived whole body sound exposure at proper water depth in tank.

[2] Cardiovascular and acid base data not available for 2 animals.

[3] Abnormal condition of the lung consisted of edema in all such cases.

Sound Exposures

Pig #	Weight (lbs)	Frequency (Hz)	Level (dB re 1 μPa)	Exposure Time (min)	Gross Pathology	Histologic Pathology
1211	107	100	161	5	Normal	Normal
		150	166	5		
		200	167	5		
		250	164	5		
		300	175	5		
		400	162	5		
1212	99	100	161	5	Normal	Normal
		150	166	5		
		200	167	5		
		250	164	5		
		270	168	5		
		300	175	5		
		400	162	5		
1213	97	270	177	5	Focal areas mottled red to purple.	Acute broncho-pneumonia present prior to experiment.
		270	177	5		
		270	177	5		
		270	177	5		
		270	177	5		
		270	177	5		

Results from Exposure Study

1. There was no detectable damage to the lungs from the sound exposure at the frequencies and levels used on the 3 animals.

2. This is consistant with a previous study at lower frequencies and similar sound pressure levels.

1. There was no detectable damage to the lungs from the sound exposure at the frequencies and levels used on the 3 animals.

2. This is consistent with a previous study by Duykers and Percy at lower frequencies (40 to 80 Hz) and similar sound pressure levels.

SLIDE 22 - Unresolved Issues

1. Depth dependence of resonance. The head out to 10 ft depth change for the study on lung resonance in humans was not adequate to characterize the effects of depth on resonance frequency. Although 10 ft of water represents a 33 % change in ambient pressure, the simplest gas resonator model predicts a change of only 15 % in the resonance frequency. It would be difficult to detect a change of this magnitude from the existing data.

2. Damage threshold as a function of frequency. There was no detectable damage from the exposure conditions used with the small number of test subjects. Therefore, it is impossible to determine damage thresholds.

3. Damage mechanisms. The assumption was made that interstitial or alveolar edema would be the most sensitive indicator of damage. This assumption was never tested nor was it understood how sound could cause edema.

Unresolved Issues

1. Depth dependence of resonance.
2. Damage thresholds as a function of frequency.
3. Damage mechanisms.

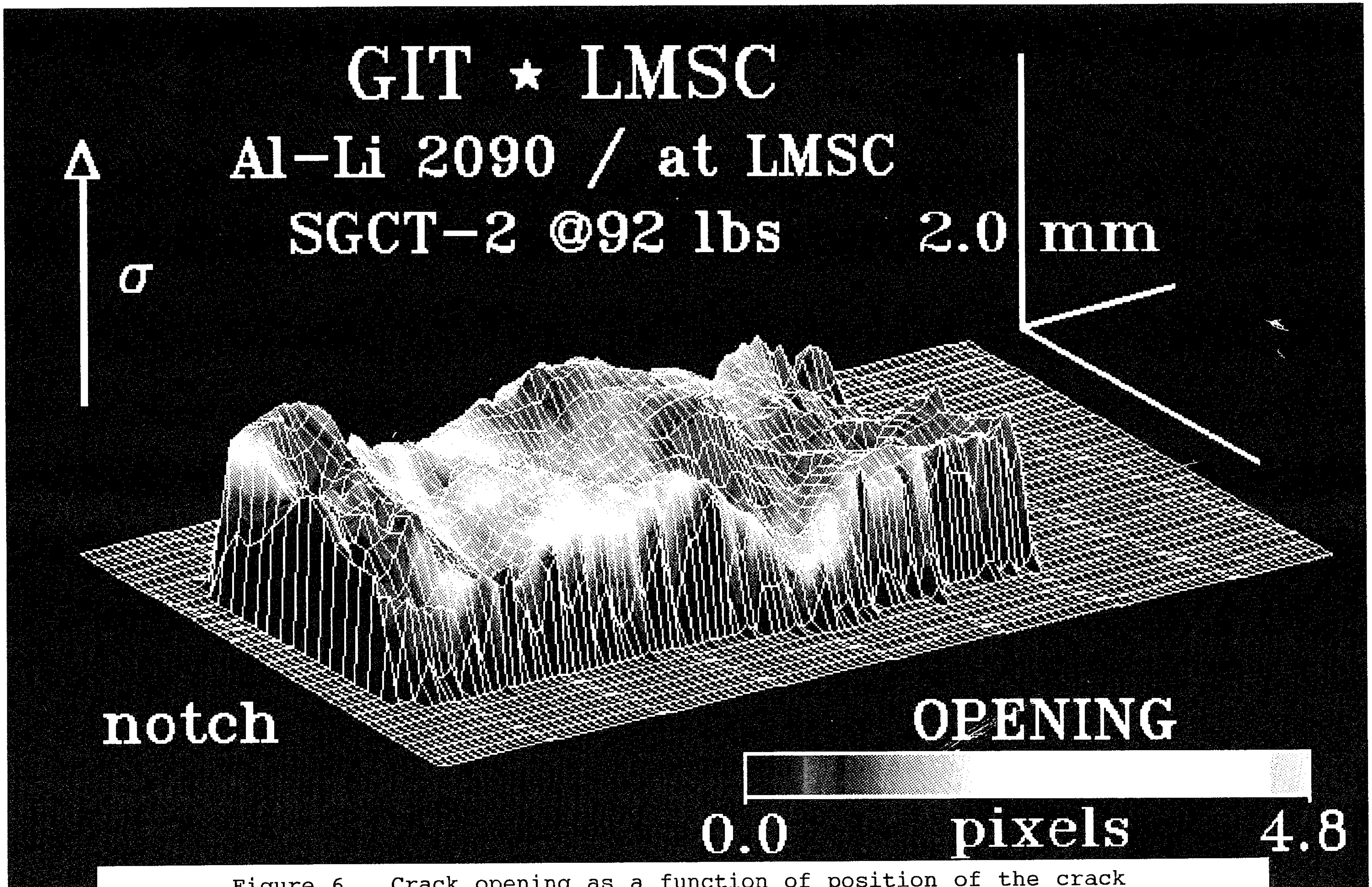


Figure 6. Crack opening as a function of position of the crack faces for the maximum load on sample CT-2. The contour lines delineate position, and the color progression black, red, blue, green and white denotes increasing opening.